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RESEARCH

Performance of tropical and subtropical maize inbred lines under well-watered and drought-stressed environments

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Abstract Drought is a major constraint for maize production in sub-Saharan Africa. Developing high-yielding drought-tolerant maize germplasm will safeguard maize yields in the ever-increasing fluctuating rainfall conditions. This study aimed to identify high-yielding inbred lines with stable performance for utilization in hybrid production. One hundred eighty-two (182) maize inbred lines were evaluated under well-watered and drought-stressed conditions at Ukulinga, Makhathini, and Cedara research stations in KwaZulu-Natal, South Africa. The experiments were carried out in a 13 × 14 alpha lattice design with two replications. The inbred lines exhibited significant differences ($p \leq 0.001$) for grain yield and yield-related traits under well-watered and drought-stressed environments. The GGE biplot identified three mega-environments, clearly separating drought-stressed from well-watered environments. Inbred lines TZISTR1190, TZISTR1231, TZISTR1261 and CML540 were superior under well-watered conditions, while TZISTR1164 and CML390 performed well under drought condition. TZISTR1190 displayed both high average yield and stability across environments. Inbred lines combining stable high yielding performance in optimum and stress conditions such as TZISTR1190 and TZISTR1231, can be incorporated into local maize breeding pipelines to develop stable high yielding resilient hybrids.

Keywords Maize · Inbred lines · Drought · Path analysis · Genotype × environment interaction

Introduction

Maize (*Zea mays* L.) is the most important cereal crop in sub-Saharan Africa (SSA). In South Africa, maize is a main staple food and a raw material for industrial products (Erenstein et al. 2022). South Africa is ranked among the top ten global leading maize-producing nations (Bradshaw et al. 2022). Nonetheless, more than 10 million people in South Africa face high levels of acute food insecurity and require immediate relief to reduce food gaps (Masipa 2017; StatsSA 2023). The Kwa-Zulu Natal Province is already considered to be in a food crisis, whereas eight of South Africa's nine provinces, Mpumalanga, Limpopo, Gauteng, North-West, Free State, Northern

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Cape, Eastern Cape, and Western Cape require measures to safeguard livelihoods (IPC 2021). At the heart of South Africa's worsening food security crisis are exorbitant food prices, which is exacerbated by the effects of climate change (Mbow et al. 2020). The price of maize has more than doubled since 2015, driven by recurrent droughts (Ala-Kokko et al. 2021).

The extent to which climate change has affected local maize yields is unknown, but Simanjuntak et al. (2023) showed that the yield losses were associated with drought events (explaining 25% of maize yield variability) nationwide. The same study suggested that heatwaves caused a 35% variability in maize yield in the Free State, while in the North-West province, a 46% variability in maize yield was due to the combination of drought and extreme precipitation. Similarly, Chukwudi et al. (2021) reported that heat stress exerted a depressive effect on maize growth and yield attributes with a potential 78% reduction in the maize harvest index. Hence, maize in Southern Africa has been identified as one of the most important crops requiring extensive climate adaptation (Lobell et al. 2008; Nhamo et al. 2019).

The introduction of exotic alleles from maize bred for tropical and subtropical environments can enhance the resilience of South African maize germplasm usually considered to be temperate. Most tropical and subtropical maize germplasm have endured several selection cycles in severe heat and drought stress environments that characterize the SSA ago-ecologies. The International Maize and Wheat Improvement Center (CIMMYT), in collaboration with the International Institute of Tropical Agriculture (IITA), have been leading in developing and deploying improved climate-resilient maize germplasm for tropical/subtropical environments. The collaboration among these two centres with the national agriculture research systems in SSA provides a large-scale, regional testing network that allows for a greater selection intensity for stress tolerance. For example, Drought Tolerant Maize for Africa (DTMA), Water Efficient Maize for Africa (WEMA), Stress Tolerant Maize for Africa (STMA) and Accelerating Genetic Gain in (AGG) projects jointly run by CIMMYT and IITA, produced several inbred lines and hybrids released throughout the continent (Shiferaw et al. 2014; Prasanna et al. 2020; Bentley 2022). These projects mainly focused on combined drought and heat tolerance by screening maize germplasm under induced drought stress with elevated temperatures. Setimela et al. (2018) reported a 20–25% yield advantage of CIMMYT'S climate resilient maize over commercial varieties in eastern and southern Africa from on-farm trials established under low-input and drought stress conditions.

Considering the substantial genetic gain for multiple stress tolerance reported by the CIMMYT and IITA programs, it can be assumed that introgression of tropical/sub-tropical germplasm in South Africa temperate maize can enhance the wide adaptation. In addition to heat and drought stress, most temperate germplasms show susceptibility to tropical diseases such as maize streak virus (MSV), Northern maize leaf blight (*Exserohilum turcicum*) and grey leaf spot (*Cercospora zea-maydis*) (Kraja et al. 2000; Ndoro et al. 2022; Dube et al. 2023). However, integration of resistance would require several selection cycles before testcross performance evaluations. Challinor et al. (2016) indicated that current global warming will decimate yields unless maize breeding and seed systems adapt forthwith. In contrast, the publicly available tropical inbred lines have already been subjected to multiple selection cycles for multiple stress tolerance. However, the direct utility of these exotic lines in local breeding programs would be restricted by numerous challenges associated with adaptability.

Prior to hybrid development, it is essential to understand the genetic diversity present within the chosen inbred lines, particularly concerning traits related to grain yield and stress tolerance. Additionally, assessing the breeding value of these inbred lines is crucial to inform the selection of parental candidates for the desired hybrids (Fritsche-Neto et al. 2021). Hence in this study, we evaluated a diverse set of inbred lines from CIMMYT and IITA's tropical and subtropical breeding programs for their performance in South Africa.

The additive main effect and multiplicative interaction (AMMI) model (Annicchiarico 1997), genotype and genotype × environment (GGE) biplot Yan and Kang (2002), and joint regression analyses Eberhart and Russell (1966) are the most common methods used for stability studies commonly referred to as genotype × environment interaction (GEI) effects. The complex relationship between genotype performance and environmental factors poses challenges in identifying superior genotypes across multiple environments (Mushayi et al. 2020). The existence of GEI decreases the correlation between genotype and phenotype, limiting the rate of progress resulting from selection (Chalwe et al. 2017). Therefore, it is imperative to examine the nature and extent of the G×E on the performance of tropical and subtropical inbred lines in South Africa. Mushayi et al. (2020) suggests the potential of exotic germplasm as sources of adaptability genes for temperate germplasm. However, there is limited information on the performance of tropical and subtropical germplasm in temperate regions. In this study we explored the adaptability of tropical and subtropical germplasm in the South African warm temperate environments to select potential testers for future test cross performance evaluations with temperate lines.

Materials and methods

Plant materials and study sites

Planting material comprised of one hundred eighty-two (182) diverse maize inbred lines sourced from the International Maize and Wheat Improvement Centre (CIMMYT), the International Institute of Tropical Agriculture (IITA), and the University of KwaZulu-Natal (UKZN). The inbred lines from CIMMYT and IITA were specifically selected for their resilience against various biotic and abiotic stresses. The UKZN inbred lines represented a subset of commercial germplasm. A detailed description of the germplasm utilized in this study is presented in Table S1.

The study was conducted across three different locations in the KwaZulu-Natal province of South Africa, namely Ukulinga (29.67 S; 30.41 E; 809 masl.), Cedara (29.76 S; 30.26 E; 1068 masl.) and Makhathini (27.39 S; 32.10 E; 77 masl.) research stations. The Makhathini site is characterized as subtropical low altitude dry environment and represents the drought stress conditions. Ukulinga and Cedara research stations are characterized as subtropical medium altitude environments of high rainfall and represent well-watered growing conditions. Each season and site combination gave unique environmental conditions due to variations in temperature and rainfall. Therefore, due to site × season combinations, a total of five environments were used for evaluating the inbred lines. The conditions prevailing in Makhathini during the 2021 and 2022 seasons were considered as environment 1 (E1) and 2 (E2), respectively. Cedara in the 2021 season was referred to as E3, while Ukulinga was considered E4 in 2021 and E5 in 2022 seasons. The geographic location, altitude, and prevailing weather conditions of the study locations are presented in Table 1.

Experimental design and management

The experiments at each location were planted in a 13 × 14 alpha lattice design with two replications. Each inbred line was planted in a 5 m long row, with intra and inter-row spacing of 0.8 m and 0.3 m, respectively, yielding a plant population density of 41 6666 plants per hectare. Two seeds were planted per hill and thinned to one, two weeks after planting. The field management adhered to the prescribed practices for maize production. Weed control was done using pre-and post-emergence herbicides, as well as occasional mechanical weeding. Supplementary overhead irrigation was administered as needed in all experimental trials throughout the initial vegetative

Table 1 Geographical coordinates and environmental conditions for the study sites

Sites	Environments	Latitude	Longitude	Altitude (masl.)	Rainfall (mm)	Min temp (^{o}C)	Max temp (^{o}C)	Management
Makhathini 21	E1	27.39 S	32.10 E	77	500	10	32	Drought
Makhathini 22	E2	27.39 S	32.10 E	77	588	16	34	Drought
Cedara 21	E3	29.76 S	30.26 E	1068	697	10	24	Well-watered
Ukulinga 21	E4	29.67 S	30.41 E	809	676	14	25	Well-watered
Ukulinga 22	E5	29.67 S	30.41 E	809	738	18	30	Well-watered

phases. At the Makhathini research station, the inbred lines were subjected to drought stress through the discontinuation of irrigation for two weeks before anthesis. Irrigation was then applied for 3 h when soil moisture reached 35% field capacity during grain filling.

Data collection

Data were recorded on days to anthesis (DA), days to silking (DS), plant height (PH), ear height (EH), ear length (EL), ear diameter (ED), kernel rows per ear (KR), kernels per row per ear (KRE), field weight (FW), grain moisture (MOI), and grain yield (GY). Grain yield (GY) expressed in t ha^{-1} was obtained from grain weight per plot adjusted to 12.5% grain moisture following CIMMYT (1999).

$$GY = \left(\frac{GW}{NP}\right) * 10 * \left(\frac{100 - MO}{87.5}\right)$$

where: GY = Grain yield (kg/ha), GW = Grain weight at harvest (kg/ha), MO = Moisture content (%) of grains at harvest. 87.5 = Standard dry matter of grain at 12.5% as required by the maize grain market authorities in South Africa, NP = Net plot area (number of rows × intra-row spacing × number of stations × inter-row spacing).

Data analysis

Separate analyses of variance (ANOVA) were performed on plot means for all collected data under well-watered and drought-stressed conditions using R software (R Core Team 2023), excluding three inbred lines which had poor plant stand in the stress treatment. Trait BLUPs were computed using DeltaGen (https://www.deltagen.agr.nz). The regression coefficient (*b*) was used as a stability response parameter of the joint regression analysis. Inbred lines with a slope coefficient $b < 0.7$ were considered unresponsive to different environments, *b* coefficient ranging between 0.7 and 1.3 referred to average stability, while inbred lines with $b > 1.3$ signalled responsiveness to G×E (Sudarić et al. 2006). Path coefficient analysis of grain yield and yield components was computed following the methodology described by Dewey and Lu (1959).

The grain yield BLUPs were used for the GGE biplot analysis to assess the stability of grain yield and the response patterns of the inbred lines across environments (Yan and Kang 2002). The analysis was done using the singular value decomposition centralized on inbred line and environment as follows:

$$\hat{Y}ij = \mu + \beta j + \lambda 1\xi i1\eta 1j + \lambda 2\xi i2\eta 2j + \epsilon ij$$

where $\hat{Y}ij$ s defined as the expected yield of the *i*-th inbred line in the *j*-th environment, μ is the grand yield mean, βj is the main effect of the *j*-th environment, λ1 and λ2 are the singular values of the first two principal components, PC1 and PC2, respectively; ξi and $\xi 2$ are the eigenvectors of the i-th inbred line for PC1 and PC2, respectively; whereas $\eta 1j$ and $\eta 2j$ are the eigenvectors of the j-th environment for PC1 and PC2, respectively; and ϵij is the residual that cannot be explained by G or GE effect.

Results

Analysis of variance of maize grain yield and yield-related traits of the inbred lines under well-watered, drought-stressed, and across test environments

Separate analyses of variance revealed significant differences ($p < 0.001$) across the test inbred lines for all evaluated traits, both under well-watered and drought-stressed conditions (Table 2). Under well-watered conditions, the environmental effects exhibited significant differences for all traits except for DA. While under drought-stressed conditions no significant differences were observed for DA and DS. The genotype by environment interaction effects under well-watered conditions showed significant differences among the genotypes for PH, EH, FW, and GY. On the contrary, only EH and ED ($p < 0.01$) showed significant differences for genotype by environment interaction effects under drought-stressed conditions. The combined analysis of variance across stressed and optimal conditions revealed highly significant differences ($p < 0.001$) among inbred lines for all the studied traits (Table 3). Similarly, the environment (E) and the environment by genotype interaction (GEI) showed significant differences ($p < 0.001$) for all the studied traits.

Table 2 Analysis of variance for eleven agronomic traits of 182 maize inbred lines evaluated under well-watered and non-stressed conditions

Source	DF	DA	DS	PH	EH	EL	ED	KR	KRE	FW	MOI	GY
Well-watered conditions												
TRT	181	121.8***	115.9***	6761***	3216.6***	36.1***	190.7***	22.5***	192.0***	2.5***	13.8***	8.5***
ENV	2	3.0	282.0***	53039***	5865.0***	59.6***	1977.3***	31.6***	290.9***	28.1***	780.1***	71.9***
REP	1	114.7**	377.4***	7699***	5252.2***	4.5	371.1***	12.7*	9.3	8.3***	190.5***	17.9***
TRT:ENV	362	14.0***	14.8	309***	126.6***	4.1	34.5	3.7*	20.5	0.4***	2.5	0.9***
ENV.REP	2	21.7	73.8*	51534***	1080.8***	11.3	58.4	9.4	34.2	3.8***	259.6***	1.7
REP.BLK	6	37.2*	37.7*	1803***	1225.3***	13.3*	134.2***	8.5*	70.2**	2.1***	2.9	4.1***
Residual	537	16.7	15.9	386	136.4	3.5**	32.11	3.1	19.8	0.2	53.4	0.6
Drought-stressed conditions												
TRT	181	821.1***	818.0***	5260.7***	1201.0***	28.5***	350.5***	24.5***	132.4***	1.1***	22.4***	5.3***
ENV	1	0.4	0.3	2939.9***	2182.0***	133.7***	1461.8***	4.9	536.0***	23.1***	445.2***	97.0***
REP	1	117.1	80.5	1530.9***	493.8***	79.4***	117.1	24.0**	2.6	0.1	391.7***	0.0
TRT:ENV	181	42.4	41.0	112.7	9.7	0.5	3.3	0.4	1.3	0.0	1.3	1.1
ENV.REP	1	168.3	162.6	248.4*	21.2*	0.7	3.5*	0.8	1.8	0.0	0.9	0.0
REP.BLK	6	77.4	73.6	1391.0***	904.6***	15.0***	432.0***	5.3	33.7*	0.8***	26.6***	3.6
Residual	356	48.9	47.5	357.7	60.2	2.8	36.0	2.6	13.9	0.1	2.3	0.5

*, **, and *** denote significant difference at $P < 0.05$, $P < 0.01$ and $P < 0.001$, respectively, *DF* degrees of freedom, *DA* days to anthesis, *DS* days to silking, *PH* plant height, *EL* ear length, *ED* ear diameter, *KR* kernel rows per ear, *KRE* kernel per row per ear, *FW* field weight, *MOI* grain moisture, and *GY* grain yield

Table 3 Combined analysis of variance for eleven agronomic traits of 182 maize inbred lines evaluated under well-watered and non-stressed conditions across five environments

Source	DF	DA	DS	PH	EH	EL	ED	KR	KRE	FW	MOI	GY
Combined well-watered and drought-stressed environments												
TRT	181	397.8***	396.6 ***	6313***	2353***	35.6***	260.1***	22.9***	166.4***	2.0***	18.2***	7.5***
ENV	4	2792.9***	3629.5***	81458***	54680***	84.3 ***	2633.6***	20.2***	514.0***	23.6***	544.6***	69.9***
REP	1	229.3**	429.4***	8596 ***	1771***	15.9*	473.7***	34.3***	1.8	4.3***	3.3	10.7***
TRT:ENV	724	153.9***	151.9***	1653***	582***	9.4***	88.3***	8.0***	50.1***	0.6***	6.1***	2.1***
ENV.REP	4	53.6	84.7*	25988***	1539***	22.8***	33.7	5.5	20.1	2.9***	274.7***	2.6**
REP.BLK	6	22.3	22.0	1797***	1090***	18.8***	259.6***	7.2*	41.8*	1.2***	12.4***	3.61***
Residual	899	30.0	28.9	381	112	3.3	35.4	3.0	117.8	0.2	3.1	0.6

*, **, and *** denote significant difference at $P < 0.05$, $P < 0.01$ and $P < 0.001$, respectively, *DF* degrees of freedom, *DA* days to anthesis, *DS* days to silking, *PH* plant height, *EL* ear length, *ED* ear diameter, *KR* kernel rows per ear, *KRE* kernel per row per ear, *FW* field weight, *MOI* grain moisture, and *GY* grain yield

Mean performance of the inbred lines under drought-stressed, well-watered conditions and across test environments

The trait BLUPs for the evaluated inbred lines are represented in Table S2. The mean grain yield was higher under well-watered conditions with inbred lines CML540 (5.9 t ha^{-1}), TZISTR1261 (5.6 t ha^{-1}), and TZISTR1164 (5.5 t ha^{-1}) recording the highest grain yields. While under drought-stressed conditions, 18 UK1-54 (5.4 t ha^{-1}), TZISTR1164 (5.3 t ha^{-1}), and CML440 (5.0 t ha^{-1}) recorded the highest grain yield. Plant and ear height were significantly reduced under drought conditions with an average of 164.0 cm, and 58.8 cm, respectively. Whereas, under well-watered conditions, plant and ear height were 182 and 79 cm, respectively. The grain yield of the 20 best-performing inbred lines across the 5 environments is presented in Table 4. The overall mean yield across the 5 environments of these inbred lines ranged from 3.68 to 4.75 t ha^{-1}. Inbred lines G1 (TZISTR1190), G2 (TZISTR1231), and G3 (TZ-14) had the highest grand mean yield of 4.75, 4.50, and 4.49 t ha^{-1}, respectively across the environments. Environment (E4) had the highest grand mean of 5.08 t ha^{-1} and the inbred lines CML540 (6.34 t ha^{-1}), TZISTR1261 (6.22 t ha^{-1}), and CML548 (5.53 t ha^{-1}), were noteworthy. However, TZISTR1261 recorded the lowest grain yield in E1 and E2 of 0.86 and 1.35 t ha^{-1}, respectively. Whereas it was among the highest-yielding inbred lines in E3, E4, and E5 with grain yield of 5.28, 6.22, and 5.33 t ha^{-1}, respectively.

The regression coefficients (*b* values) were used to evaluate the stability of the best 20 inbred lines across the 5 environments (Table 4). The *b* coefficients varied between −1.82 (G19) to 3.36 (G4). For genotypes to be considered stable, the *b* value should ideally be 1.0 or greater than the general average. According to the observed *b* values, the inbred line G1 (TZISTR1190) showed the highest grain yield (4.75 t ha^{-1}), with a regression coefficient of 1.21.

Path coefficient analysis of grain yield and yield-related traits under well-watered and non-stressed conditions

Under well-watered conditions, DA (0.01) showed a positive direct effect on grain yield (Table 5), and

Table 4 Mean grain yield (t ha^{-1}) and regression coefficients (*b*) of top 20 inbred lines across 5 environments

Inbred codes	Inbred lines	E1	E2	E3	E4	E5	Across	*b* value
G1	TZISTR1190	3.42	4.08	5.90	5.48	4.47	4.75	1.21
G2	TZISTR1231	3.42	3.95	4.71	5.04	5.20	4.50	0.75
G3	TZ-14	5.25	5.81	3.53	4.33	3.56	4.49	−1.08
G4	CML540	1.10	2.05	4.78	6.34	5.11	4.41	3.36
G5	TZISTR1275	2.46	3.46	4.02	5.01	6.09	4.20	1.63
G6	TZ-45	3.08	3.91	3.85	5.10	4.63	4.17	1.06
G7	18 UK1-24	3.76	4.75	3.27	5.40	3.52	4.07	0.40
G8	CML571	1.51	2.49	5.47	5.45	5.32	4.05	2.56
G9	TZI3STR	3.62	4.61	3.51	5.11	3.07	4.01	0.09
G10	CLHP0156	2.81	3.45	4.05	5.02	4.42	3.95	1.03
G11	TZ-11	2.36	3.34	3.86	5.67	4.68	3.95	1.85
G12	CLHP0113	3.09	3.77	3.73	5.12	3.66	3.93	0.82
G13	TZSTRI112	2.27	2.91	4.51	5.44	4.24	3.92	1.83
G14	CML541	3.53	4.37	3.44	4.94	3.61	3.89	0.26
G15	TZ-34	2.90	3.89	3.67	4.77	4.03	3.88	1.00
G16	CML548	1.90	2.64	4.50	5.53	4.96	3.86	2.04
G17	TZISTR1261	0.86	1.35	5.28	6.22	5.33	3.83	3.34
G18	TZ-8	3.93	4.69	3.29	3.87	2.96	3.75	−0.48
G19	TZISTR1164	4.97	5.70	2.35	2.93	1.96	3.70	−1.82
G20	CML390	3.42	4.17	3.19	4.83	2.95	3.68	0.14
Mean		2.98	3.77	4.05	5.08	4.19	4.05	

Table 5 Direct and indirect effects of secondary traits on grain yield of maize inbred lines under well-watered conditions

	Well-watered conditions										
	DA	DS	PH	EH	EL	ED	KR	KRE	FW	MOI	GY
DA	**0.01**	−0.03	0.00	0.00	−0.01	0.00	0.00	0.01	−0.06	−0.02	**−0.09**
DS	0.01	**−0.03**	0.00	0.00	−0.01	0.00	0.00	0.01	−0.08	−0.01	**−0.12**
PH	0.00	0.00	**0.03**	−0.03	0.03	0.00	0.01	−0.02	0.64	−0.01	**0.64**
EH	0.00	0.00	0.03	**−0.03**	0.02	0.00	0.00	−0.02	0.60	−0.01	**0.59**
EL	0.00	0.00	0.02	−0.02	**0.04**	0.00	0.01	−0.03	0.63	−0.01	**0.64**
ED	0.00	0.00	0.02	−0.02	0.02	**0.00**	0.01	−0.02	0.55	−0.01	**0.57**
KR	0.00	0.00	0.01	−0.01	0.01	0.00	**0.02**	−0.01	0.31	−0.01	**0.33**
KRE	0.00	0.00	0.02	−0.02	0.03	0.00	0.01	**−0.03**	0.68	−0.01	**0.69**
FW	0.00	0.00	0.02	−0.02	0.03	0.00	0.01	−0.02	**0.93**	−0.01	**0.93**
MOI	0.00	−0.01	0.01	−0.01	0.01	0.00	0.00	−0.01	0.30	**−0.03**	**0.27**

DA days to anthesis, *DS* days to silking, *PH* plant height, *EL* ear length, *ED* ear diameter, *KR* kernel rows per ear, *KRE* kernel per row per ear, *FW* field weight, *MOI* grain moisture, and *GY* grain yield

Diagonal bold represent the direct effects; vertical bold is the correlation coefficient with grain yield

it also illustrated a negative indirect effect with DS (−0.03), EL (−0.01), FW (−0.06), and MOI (−0.02). Plant height (0.03) exhibited a positive direct effect on grain yield and further displayed a strong indirect effect on FW (0.64). Field weight (0.93) recorded the highest positive direct effect on grain yield and had a positive direct effect on PH (0.02), EL (0.03), and KR (0.01). Under drought-stressed conditions, DA (0.01) displayed a positive direct effect on grain yield and showed a negative indirect effect with DS (−0.04)

Table 6 Direct and indirect effects of secondary traits on grain yield of maize inbred lines under drought-stressed conditions

	Drought-stressed conditions										
	DA	DS	PH	EH	EL	ED	KR	KRE	FW	MOI	GY
DA	**0.01**	−0.04	0.00	0.00	0.00	0.00	0.00	0.00	−0.17	0.00	**−0.17**
DS	0.01	**−0.04**	0.00	0.00	0.00	0.00	0.00	0.00	−0.15	0.00	**−0.14**
PH	0.00	−0.01	**0.01**	0.01	0.00	0.01	0.00	0.00	0.49	0.00	**0.55**
EH	0.00	−0.01	0.01	**0.01**	0.00	0.01	0.00	0.01	0.46	0.00	**0.57**
EL	0.00	0.00	0.00	0.01	**0.00**	0.01	0.00	0.00	0.68	0.00	**0.58**
ED	0.00	0.00	0.00	0.00	0.00	**0.03**	0.01	0.01	0.40	0.00	**0.57**
KR	0.00	0.00	0.00	0.00	0.00	0.01	**0.02**	0.03	0.37	0.00	**0.41**
KRE	0.00	0.01	0.01	0.01	0.00	0.01	0.01	**0.03**	0.63	0.00	**0.73**
FW	0.00	0.00	0.01	0.01	0.00	0.02	0.01	0.01	**0.93**	0.00	**0.99**
MOI	0.00	−0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.10	**−0.02**	**0.22**

DA days to anthesis, *DS* days to silking, *PH* plant height, *EL* ear length, *ED* ear diameter, *KR* kernel rows per ear, *KRE* kernel per row per ear, *FW* field weight, *MOI* grain moisture, and *GY* grain yield

Diagonal bold represent the direct effects; vertical bold is the correlation coefficient with grain yield

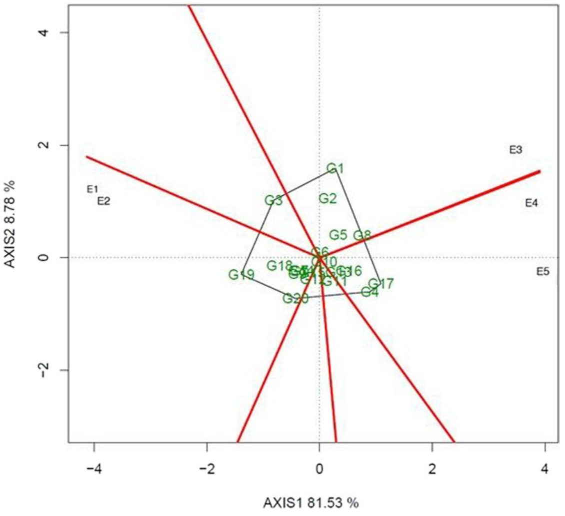

Fig. 1 GGE biplot showing which-won-where pattern of the 20 maize inbred lines evaluated across 5 environments [E1and E2 (Makhathini), E3 (Cedara), E4 and E5 (Ukulinga)]

and FW (−0.17). Field weight (0.93) exhibited a strong positive direct effect on grain yield (Table 6).

GGE biplot analysis

The GGE biplot analysis was used to visually assess the influence of genotype × environment interaction effect on maize grain yield among the top 20 best-performing inbred lines across the environments. A which-won-where biplot, was constructed to determine the performance of different inbred lines in specific environments and identifying the inbred lines that excelled in a particular environment or a group of environments (Fig. 1). The lines that extend perpendicularly from the origin of the biplot, are referred to as sectors. These sectors provide a clear visual representation of the inbred lines that exhibit superior performance in each environment. A polygon was constructed by connecting inbred lines that were furthest from the biplot origin, ensuring that all inbred lines were contained inside the polygon. The G1 (TZISTR1190), G17 (TZISTR1261), G20 (CML390), G19 (TZISTR1164), and G3 (TZ-14) delimited the polygon forming the vertex inbred lines with the longest vectors. Therefore, these inbred lines exhibited a higher degree of responsiveness to the environment in their respective orientations as compared to other inbred lines such as G2 (TZISTR1231), G5 (TZISTR1275), G11 (TZ-11), G13 (TZSTRI112), and G14 (CML541) contained within the polygon.

The polygon was partitioned into six sectors, and environments contained inside each sector are referred to as mega-environments. However, only sectors containing one or more environments qualify as mega-environments. Therefore, in this study, the GGE-biplot grouped the testing sites into three mega-environments. Environment E3 (Cedara) formed the first mega-environment, E4 and E5 (Ukulinga) formed the second mega-environment, whereas E1 and E2 (Makhathini) formed the third mega-environment. G1 (TZISTR1190) and G2 (TZISTR1231) were the best-performing inbred lines in 1st mega-environment, E3. In the 2nd mega-environment comprising E4 and E5, G4 (CML540), and G17 (TZISTR1164) were the best-performing inbred lines. However, the 3rd mega environment containing E1 and E2, had G19 (TZISTR1164), and G20 (CML390), as the best-performing inbred lines.

Mean vs stability

The average environment coordination (AEC) view of the GGE biplot depicts the mean performance and stability classification of the inbred lines across five environments (Fig. 2). The study revealed high-performing and stable inbred lines through the drawing of an AEC on the genotype-focused biplot. The directional arrow serves as an indicator of superior inbred lines that demonstrate good performance across various environments, while the intersecting lines represent increased variability, which implies stability in both directions. The G1 (TZISTR1190), G2 (TZISTR1231), G8 (CML571) and G5 (TZISTR1275) were the stable highest yielding inbred lines, whereas G17 (TZISTR1261), G19 (TZISTR1164), and G20 (CML390)) were the lowest yielding inbred lines. Most inbred lines showed moderate and stable yield performance across the environments. However, G3 (TZ-14), G17 (TZISTR1261), and G19 (TZISTR1164) were the most unstable inbred lines.

Ranking of inbred lines

The inbred lines ranking can detect an ideal inbred line in contrast to other inbred lines evaluated. The inbred lines that are considered ideal exhibit both high average yields and consistent performance across various environments. These inbred lines are situated at the center of the concentric circle and exhibit no genotype by environment interaction and are represented by a small distance between the genotype average environment axis (AEA). The greater the distance between the inbred line and the concentric circle, the less ideal is the inbred line. The inbred lines G1 (TZISTR1190) was positioned at the center of the concentric circle, followed by G2 (TZISTR123) situated closer to the concentric circle (Fig. 3). These two inbred lines are regarded as ideal

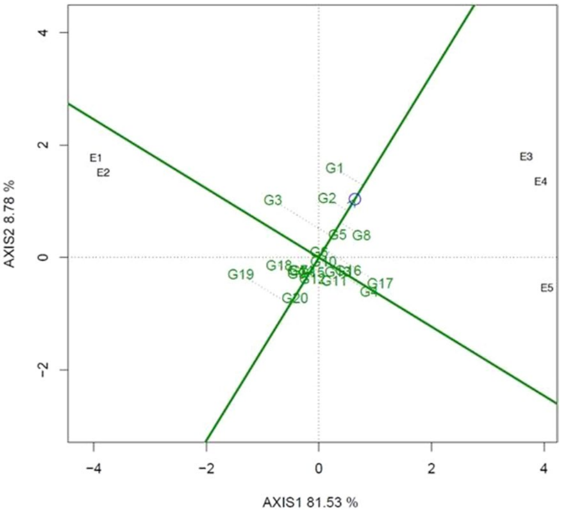

Fig. 2 Analysis of the mean performance and stability of genotypes using the GGE biplot [E1and E2 (Makhathini), E3 (Cedara), E4 and E5 (Ukulinga)]

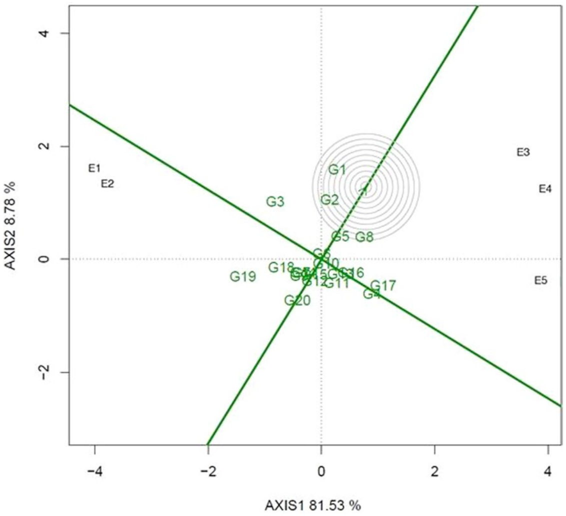

Fig. 3 GGE-biplot showing the best maize inbred lines based on mean grain yield performance and stability across 5 environments [E1and E2 (Makhathini), E3 (Cedara), E4 and E5 (Ukulinga)]

inbred lines across the test environments because of their position, indicating that they are stable inbred lines. The inbred lines G15 (TZ-34), G19 (TZISTR1164), and G20 (CML390) were the least yielding inbred lines since they are located far away from the concentric circle.

Discriminativeness vs representativeness

Selecting the most suitable testing environment contributes to the efficiency of a breeding program, and the discriminating power and representativeness view of the GGE-biplot provides an assessment of the testing environments (Fig. 4). An "ideal" testing environment not only effectively discriminates among inbred lines but also accurately represents all other environments under study, enabling it to predict the best inbred line with precision. The 'average environment' is represented by a small circle on the AEA. The cosine of the angle between any environment vector and the average environment axis approximates the correlation coefficient between the genotype values in that environment and the genotype means across the environment. Test environments making small angles with the AEA are more representative of all environments than those making large angles. Hence, E 3, E4, and E5 were the most discriminating, whereas E1 and E2 were the least discriminating environments.

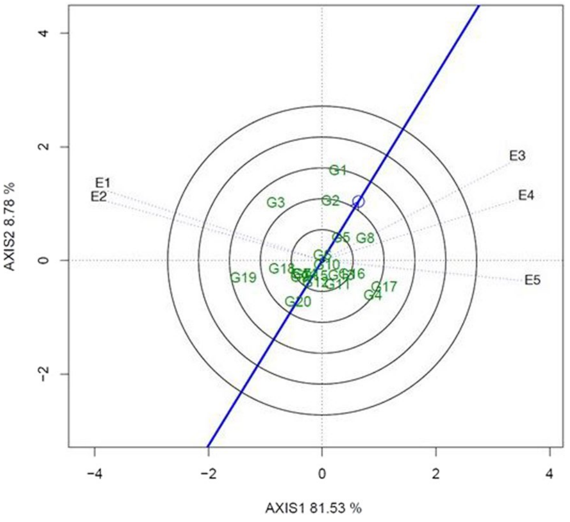

Fig. 4 The "discriminating power vs. representativeness" view of the GGE biplot based on yield data of 20 inbred lines across 5 environments [E1and E2 (Makhathini), E3 (Cedara), E4 and E5 (Ukulinga)]

Discussion

Tropical and subtropical maize germplasm can adapt to intense heat and frequent drought stress conditions that characterize SSA climates (Ndoro et al. 2022). Hence, the infusion of exotic alleles derived from maize bred for tropical and subtropical climates has the potential to bolster the resilience of South Africa's temperate maize germplasm (Dube et al. 2023). In this study, the performance of a collection of tropical and subtropical inbred lines was assessed across contrasting environments in the KwaZulu-Natal province of South Africa to select the most ideal inbreds for future breeding. The genotype-by-environment interaction effect causes changes in cultivar performance ranking across different environments, thereby impacting the efficacy of selection in a breeding program. Genotype-by-environment interactions arise due to variations in genotype sensitivity to diverse environmental conditions (Yan et al. 2007; Begna 2022).

The separate analysis of variance revealed that the effect of inbred lines was highly significant for most measured traits. The significant effect of genotypes not only demonstrates the genetic difference between genotypes but also suggests the possibility of selecting unique genotypes (Khan and Mahmud 2021; Dube et al. 2023). Furthermore, the combined analysis of variance revealed that the effect of genotypes × environment was highly significant for grain yield and other traits. The significant effect of genotypes × environment has strong implications for breeding for specific adaptation (Sserumaga et al. 2016). The Ukulinga and Makhathini sites represented subtropical, low to medium-altitude dry environments with minimal disease pressure. In contrast, the Cedara site was defined by its subtropical, medium-altitude setting characterized by high rainfall and high relative humidity, which often results in high disease pressure.

The inbred lines TZISTR1190 (4.75 t ha^{-1}), TZISTR1231 (4.50 t ha^{-1}), TZISTR1261 (3.83) and CML540 (4.41 t ha^{-1}) exhibited consistent and high grain yield performance under well-watered environments. Conversely, inbred lines, TZISTR1164 and CML390 demonstrated high yield performance specifically in E1 and E2, which represented water-stressed conditions. These results are in accordance with those reported by Twumasi et al. (2017) and Dhliwayo et al. (2009) under optimum conditions and the findings of Worku et al. (2016) under managed drought condition. One of the key priorities of both CIMMYT and IITA breeding programs is to enhance drought tolerance in maize.

Relying solely on grain yield to select drought-tolerant germplasm under drought conditions can be misleading. Therefore, it is advisable to incorporate secondary traits alongside grain yield for a more comprehensive assessment and accurate selection of drought-tolerant genotypes (Noor et al. 2019). The path coefficient values proposed by Lenka and Misra (1973) are classified as follows: negligible (0.00–0.09), low (0.10–0.19), moderate (0.20–0.29), and high (0.30–0.99). In this study under well-watered environments, days to anthesis, plant height, ear length, and kernel rows per ear exhibited partial positive direct associations with grain yield ranging between (0.00–0.04). Whereas under drought-stressed environments days to silking, and grain moisture displayed a negative direct effect on grain yield. These results are in contrast with those reported by Kinfe et al. (2015), and Pandey et al. (2017) where days to anthesis, plant height, ear length, and kernel rows per ear, exhibited high positive direct associations with grain yield. However, selection for early silking dates may improve chance of synchronisation with anthesis before critical moisture stress periods. Field weight displayed a high positive direct effect on grain yield and could be used for indirect selection to enhance grain yield in the respective environments.

The "which-won-where" biplot polygon illustrates how genotypes and environments interact, revealing crossover GEI, mega-environment differentiation, and specific adaptation patterns (Yan and Tinker 2005, 2006). In the present study, G1 (TZISTR1190), G17 (TZISTR1261) and G4 (CML 540), were the vertex and winning inbred lines in well-watered conditions. On the other hand, G19 (TZISTR1261), and G20 (CML390) were the vertex and winning inbred lines under drought stress conditions. According to Yan and Tinker (2006), the responsiveness of genotypes at the vertices of the polygon is greater in comparison to those located within the polygon. The polygon biplot was divided into six sectors, with three mega environments. If the environments fall into distinct sectors, it indicates that various genotypes performed well in different environments (Abakemal et al. 2016; Sserumaga et al. 2018). The first mega-environment (E3) contained the two highest-yielding inbred lines G1 (TZISTR1190) and G2 (TZISTR1231). The environments E4 and E5 were contained in one sector and were also characterized by high-yielding inbred lines G4 (CML540) and G17 (TZISTR1261). The third mega environment contained E1 and E2 was characterized by low-yielding inbred lines due to drought stress. However, inbred lines G19 (TZISTR1164), and G20 (CML390) exhibited high grain yield under the third mega-environment and could be considered as future testers for developing hybrids for drought tolerance. Most importantly, the mega environment analysis confirmed the existing edaphic and climatic differences which distinguished the three testing sites used in this study. We would not have been surprised if Ukulinga and Cedara research stations were classified in one mega environment because of the 30 km geographic distance between the two stations. However, the two sites offer quite diverse testing conditions because Cedara has more elevation, slightly high average rainfall, and daily temperature range. Makhathini research station is situated in the lowveld, with very high temperature range, coupled with generally low and unpredicted rainfall patterns hence it served as the drought stress environment. This means that three mega environments identified offer unique testing conditions for the breeding program.

Ideal genotypes exhibit both high mean performance and stability across environments. It is positioned at the center of the concentric circle and in the positive direction of AEA in the GGE biplot (Yan and Tinker 2006). Although an ideal genotype is uncommon and seldom manifests in practice, this concept it can be utilized as a reference for cultivar evaluation. The closer a genotype is to the concentric circle, the more desirable it is (Mitroviã et al. 2012). Therefore, in this study, TZISTR1190 and G2 TZISTR1231 were the ideal inbred lines based on their positions. These inbred lines can be utilized as testers in future tropical × temperate test cross performance tests.

An average environment coordination (AEC) method was used to evaluate the yield performance and stability of the maize inbred lines. Evaluation of inbred lines within a mega-environment should encompass both average performance and stability to prevent the effects of random GEI. In this study, TZISTR1190 and TZISTR1231 inbred lines had the highest mean yield and were more stable than other inbred lines. On the other hand, TZ-34 was stable but among the lowest yielding inbred lines. These findings suggest that the two stable superior inbred lines may be considered as broad-based testers in future testcross performance trials.

Conclusion

Our study demonstrated that the grain yield of inbred lines was higher under well-watered conditions than under drought-stressed conditions. Under well-watered conditions high grain yield was associated with tall plants and wider ear diameter, whereas under drought-stressed high grain yield was associated with shorter days to silking and anthesis and high field weight. The G1 (TZISTR1190), G17 (TZISTR1261) and G4 (CML 540) emerged as the top-performing inbred lines under well-watered conditions. While under drought stress G19 (TZISTR1261), and G20 (CML390) were the vertex and winning inbred lines. These inbred lines could be assessed for their breeding value as testers for developing hybrids for optimal and drought-stress environments.

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Author contributions All the authors participated sufficiently in the completion of this work. S.P. conducted the experiment, collected phenotypic data, and wrote the manuscript; A.I.T, F.R., and J., guided the experimental design, data collection, writing, reviewing, and editing of the manuscript.

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Data availability No datasets were generated or analysed during the current study.

Declarations

Conflict of interest The authors have no competing interests to declare that are relevant to the content of this article.

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